

## Assessment of fertility restorer gene ( $R_f$ ) in R-line and *Moricandia* based hybrid of Indian mustard using SCAR marker

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### Abstract

Sequence characterized amplified region (SCAR) markers were developed for fertility restorer gene that is useful for hybrid development in Indian mustard. Validation of primer (SCAR-3) associated with fertility restorer gene was tested in 50 restorer inbred plants (R-line) and their  $F_1$  derived from *Moricandia* CMS. This was further confirmed through observation of flowers, pollen fertility/sterility and seed set. The SCAR-3 produced a sharp band (200 bp) in restorer line and their respective hybrids. The genetic purity of the hybrid was above 90% while the restorer line showed higher impurity (36%). The results showed very high correspondence between the flower, pollen fertility/sterility and seed set with SCAR marker indicating potentiality of case specific utilization of this marker in testing hybrid seed purity.

**Key words:** CMS, fertility restorer gene, hybrid, Indian mustard, SCAR marker

A stable CMS line of *Brassica juncea* was derived from the somatic hybrid between *Moricandia arvensis* and *B. juncea* following back crosses with *B. juncea* [1]. The fertility restorer gene for this Cytoplasmic male sterility (CMS) was introgressed into *B. juncea* from *M. arvensis* [2, 3]. Hence, this CMS system has been used to explore heterosis in Indian mustard (*Brassica juncea* L. Czern & Coss). A stable *Moricandia* based CMS line either carries an unaltered or recombined mitochondrial genome. Subsequently, it was found that this  $R_f$  locus is capable to restore male fertility in *Moricandia* based CMS system. The purity of  $F_1$  hybrid seeds is traditionally assessed in a field grow-out test

(GOT), but these trials are time-consuming, labor-intensive and require large plots of land [4]. Furthermore, morphological differences between true and false hybrids of mustard are not always apparent and cannot be recognized easily, especially when the parents are genetically similar resulting in genetic impurity. Isozyme analysis has been used for purity testing [5]. However, this method is not only tissue specific but also requires selection of a suitable isozyme by screening a number of enzymes [6, 7]. Molecular markers provide a more accurate means for identifying and characterizing varieties, including hybrids [8]. Studies on seed purity and genotype identification have indicated that careful selection of SSR markers or sequence tagged sites can differentiate male sterile, fertility restorer and potential rogue donor lines in rice [9]. Consequently, markers linked to the mapped fertility restorer genes that can detect heterozygosity in male sterile and restorer lines have the potential to be more efficient than markers not linked to the restorer genes. A set of sequence characterized amplified region (SCAR) markers tightly linked with the restorer genes has therefore, been developed. Of these, SCAR-3 marker was found to be the closest to the  $R_f$  locus with a map distance of 0.6 cM in *Brassica juncea* [10]. The present study analyzed the efficiency of  $R_f$ -linked SCAR markers in detecting the contamination in restorer line and their respective hybrids.

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